A scalable system for production of functional pancreatic progenitors from human embryonic stem cells.

Journal: PLoS One

Publication Year: 2012

Authors: Thomas C Schulz, Holly Y Young, Alan D Agulnick, M Josephine Babin, Emmanuel E Baetge, Anne G Bang, Anindita Bhoumik, Igor Cepa, Rosemary M Cesario, Carl Haakmeester, Kuniko Kadoya, Jonathan R Kelly, Justin Kerr, Laura A Martinson, Amanda B McLean, Mark A Moorman, Janice K Payne, Mike Richardson, Kelly G Ross, Eric S Sherrer, Xuehong Song, Alistair Z Wilson, Eugene P Brandon, Chad E Green, Evert J Kroon, Olivia G Kelly, Kevin A D'Amour, Allan J Robins

PubMed link: 22623968

Funding Grants: Methods for detection and elimination of residual human embryonic stem cells in a differentiated cell product, Cell Therapy for Diabetes

Public Summary:
The team at ViaCyte has discovered and refined the methods for manufacturing pancreatic progenitor (pancreatic endoderm; PE) cells from human embryonic stem (ES) cells. After implant into animals, these cells secrete insulin in response to elevated glucose. They reliably cure diabetic rodents. In this report, ViaCyte scientists describe a modification of the method to manufacture PE which is more amenable to large scale production than the previously described method. Previously the cell differentiation (converting ES cells to PE cells) was performed in adherent cell culture; the new method involves making ES cell aggregates that are differentiated to PE in suspension culture.

Scientific Abstract:
Development of a human embryonic stem cell (hESC)-based therapy for type 1 diabetes will require the translation of proof-of-principle concepts into a scalable, controlled, and regulated cell manufacturing process. We have previously demonstrated that hESC can be directed to differentiate into pancreatic progenitors that mature into functional glucose-responsive, insulin-secreting cells in vivo. In this study we describe hESC expansion and banking methods and a suspension-based differentiation system, which together underpin an integrated scalable manufacturing process for producing pancreatic progenitors. This system has been optimized for the CyT49 cell line. Accordingly, qualified large-scale single-cell master and working cGMP cell banks of CyT49 have been generated to provide a virtually unlimited starting resource for manufacturing. Upon thaw from these banks, we expanded CyT49 for two weeks in an adherent culture format that achieves 50-100 fold expansion per week. Undifferentiated CyT49 were then aggregated into clusters in dynamic rotational suspension culture, followed by differentiation en masse for two weeks with a four-stage protocol. Numerous scaled differentiation runs generated reproducible and defined population compositions highly enriched for pancreatic cell lineages, as shown by examining mRNA expression at each stage of differentiation and flow cytometry of the final population. Islet-like tissue containing glucose-responsive, insulin-secreting cells was generated upon implantation into mice. By four- to five-months post-engraftment, mature neo-pancreatic tissue was sufficient to protect against streptozotocin (STZ)-induced hyperglycemia. In summary, we have developed a tractable manufacturing process for the generation of functional pancreatic progenitors from hESC on a scale amenable to clinical entry.